ÄKTA™ pure

ÄKTA pure is a flexible and intuitive chromatography system (Fig 1) for fast purification of proteins, peptides, and nucleic acids from microgram levels to tens of grams of target product. ÄKTA pure is a reliable system where hardware and UNICORN™ system control software are designed to work together with columns and chromatography media to meet any purification challenge.

ÄKTA pure is available in two versions: ÄKTA pure 25 designed for a broad range of research applications and purification tasks in a multiuser environment; ÄKTA pure 150 is well suited for optimizing resource utilization and productivity in routine large-scale preparative purification. The system supports a wide range of chromatography techniques and meets the automation requirements needed to deliver the highest purity. The system is configurable and can be upgraded at any time with a wide range of options to further increase its capabilities depending on your purification needs.

ÄKTA pure is the product of over fifty years expertise in protein research and three decades of experience in the development of ÄKTA purification systems.

ÄKTA pure offers the following benefits for you:

- Modular system design with a large range of options to allow flexibility in purification of proteins and peptides
- Customizable system that is easy to upgrade as your research needs develop
- Reliable system with components and integrated features based on the proven design of ÄKTA avant
- UNICORN 6 software provides simple, intuitive, and flexible preprogrammed method templates and total system control to simplify your job
- Predefined method settings for all GE Healthcare Life Sciences lab-scale chromatography columns

System overview

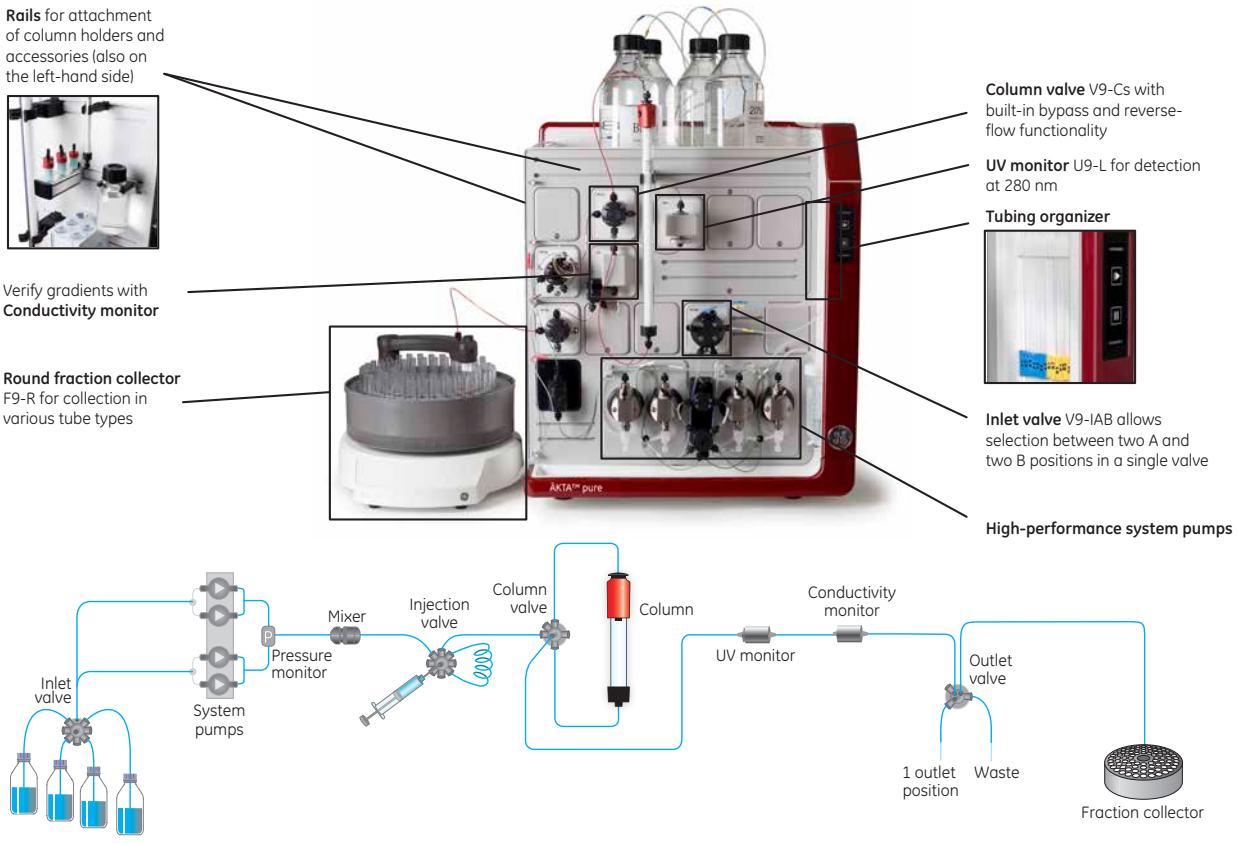
ÄKTA pure chromatography system is a highly versatile, modular system with a number of design features to facilitate reliable purification.

Fig 1. ÄKTA pure is a flexible chromatography system for the reliable purification of proteins, peptides, and nucleic acids at laboratory scale.

The system consists of the ÄKTA pure instrument and UNICORN 6 control software. The system is modular in design with all valves, monitors, and columns mounted on the forward facing wet side of the system, to allow easy interaction with the instrument modules (Fig 2). Additional components such as valves, monitors, and sensors from the wide range of optional modules can easily be added to the available positions. Multiple rails for attachment of column holders and equipment are located at the front and on the side of the instrument. A buffer tray on the top of the instrument provides a large storage area for vessels and bottles. The instrument control panel shows the system state and allows the possibility to interact with the run (pause/continue) at the touch of a button.

The system weighs only 48 kg in basic configuration and 53 kg when fully equipped with options. The relatively low weight enables easier placement in the lab and the system dimensions allow it to fit conveniently into a standard cold cabinet for work with labile samples.

(A)



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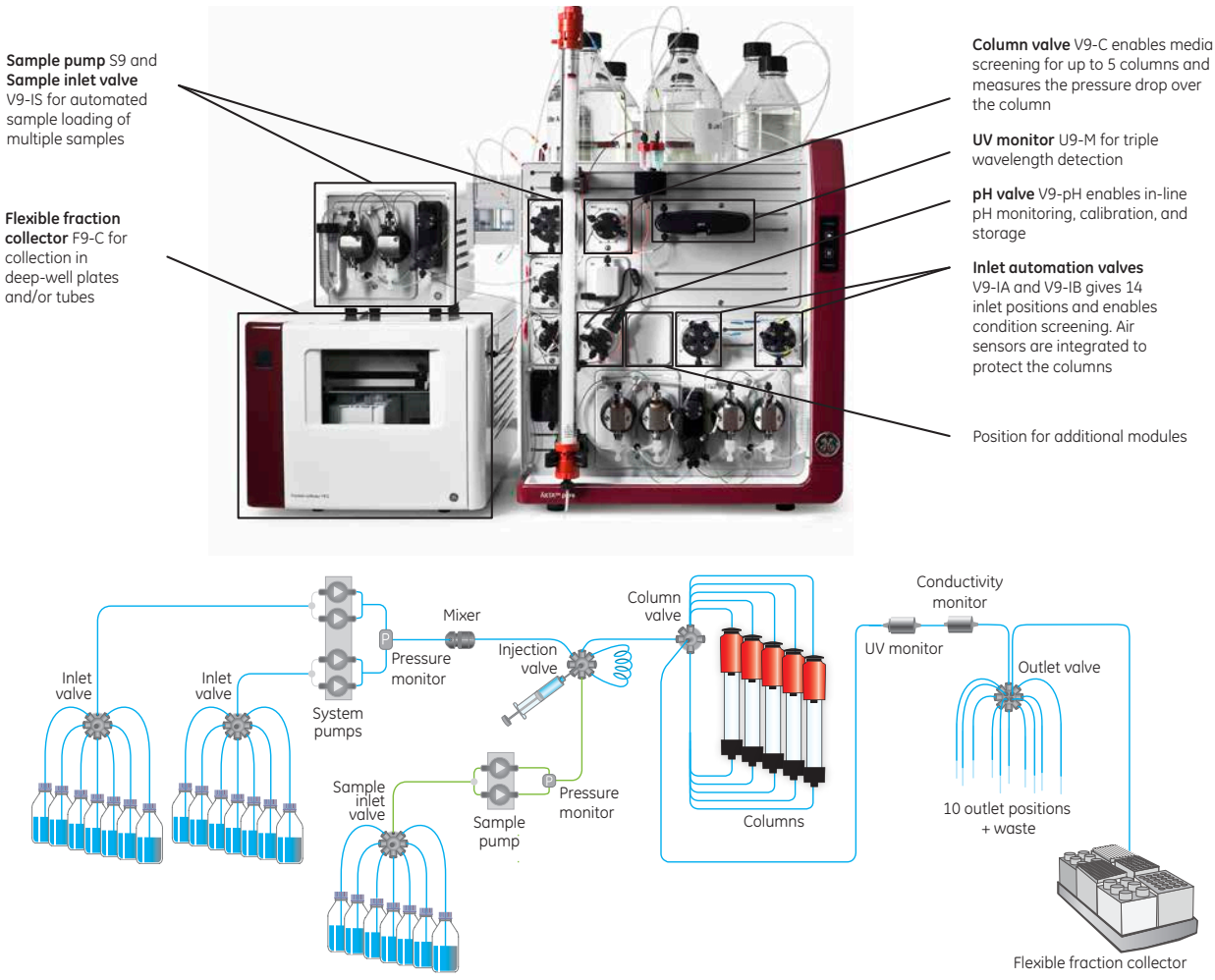


Fig 2. Two examples of system configurations for AKTA pure 25 showing positions of modules on the front panel and flow paths for each. (A) A basic system configuration for convenient protein purification. (B) System configured for high level of automation.

Regardless of configuration, ÄKTA pure always comes with two high-performance system pumps, system pressure monitor for column protection, mixer, injection valve, and UV monitor. ÄKTA pure has a wide range of optional modules to allow a large number of possibilities. The system flow path is designed to minimize band-broadening effects, and all wetted materials used in the flow path are biocompatible and resistant to commonly used solvents. The instrument front is designed with empty module positions where optional valves and monitors can be mounted to enable a flexible configuration of the flow path. Examples of two ÄKTA pure system configurations are shown in Figure 2.

UNICORN 6 control software allows a fast and easy start to creating methods and starting runs. UNICORN 6 eliminates the need for programming skills as creation of chromatography methods is done by simple drag-and-drop operations. In addition, the software is modular allowing the addition of features such as Column Logbook and Design of Experiments (DoE) functionality for method development. Licensing options for remote access to the system and/or for creating methods or evaluating results give even greater convenience. If preferred, the system can be set up so that it enters "power save mode" after method end, which enables reduction of power consumption by around 80%.

ÄKTA pure system components and available options are described in the following sections in more detail.

ÄKTA pure standard components

System pump

The two system pumps are based on the modern technology developed for ÄKTA avant systems. The robust construction delivers reproducible flow rates at both low and high back pressures, allowing short separation times.

Each pump consists of one pair of pump heads, which deliver low-pulsation flow to the mixer. The continuous and accurate flow rates generated enable reproducible isocratic or gradient elution. For ÄKTA pure 25 the system pumps provide a flow rate range of up to 25 mL/min at maximum operating pressure of 20 MPa. For ÄKTApure 150 the flow rate is up to 150 mL/min at maximum operating pressure of 5 MPa. For column packing, ÄKTA pure 25 and 150 can be used at flow rates up to 50 mL/min and 300 mL/min, respectively. A system pressure monitor is connected to the pumps to continuously measure system pressure and enable the flow rate to be automatically adjusted to avoid reaching any defined pressure limit.

Mixer

The mixer enables homogeneous buffer composition during gradient runs. The choice of mixer chamber size depends on the flow rate and buffers used, with a larger mixer volume required for higher flow rates or difficult-to-mix buffers. Table 1 shows the mixer chamber sizes available for each instrument.

Table 1. Mixer chamber sizes available

System	Mixer chamber sizes
AKTA pure 25	Included: 1.4 mL; options: 0.6 and 5 mL
AKTA pure 150	Included: 1.4 and 5 mL; option: 15 mL

An in-line filter is mounted on the mixer. The filter is easy to change, and the mixer is also easily changed by snapping it in or out of the mixer holder. The mixer size used for any given run is always noted in the result file.

Injection valve

The injection valve allows for a variety of sample application techniques using sample loops or Superloop™ sample injection device. The novel valve design eliminates the need for replumbing when changing between various sample application techniques. A sample loop with a volume of 500 µL is delivered with the system. Sample loops can be filled manually, via a syringe, or with a sample pump; the same sample application options apply to the use of Superloop. Sample loops can also be filled using the system pump.

Moreover, sample can also be applied to the column directly using an optional sample pump or the system pump.

UV monitoring

ÄKTA pure is equipped with either a fixed wavelength UV monitor or a variable multiwavelength UV and visible spectrum monitor.

The fixed wavelength (280 nm) UV monitor (U9-L) incorporates LED technology, which is durable and reliable and is ready to use at start-up. Moreover, UV monitor U9-L does not heat the sample. The monitor is available with a 2 mm flow cell as standard (included at delivery) and an optional 5 mm flow cell when higher sensitivity measurements are required.

To determine protein separation at different wavelengths, UV monitor U9-M is designed for multiwavelength detection in the UV and visible spectrum from 190 to 700 nm.
UV monitor U9-M allows monitoring of up to three wavelengths simultaneously (Fig 3 and 6). For optimized performance when purifying samples with different protein concentrations, there are three flow cell path lengths available; 0.5, 2 (included at delivery), and 10 mm. The flow cell design, together with fiber optic technology, provides a high signal-to-noise ratio without causing any local heating of the UV flow cell. The monitor contains a high-intensity xenon lamp with a long lifetime that requires minimal start-up time. Every time the instrument is switched on, the monitor is automatically calibrated. All U9-M UV cells are calibrated at manufacturing. The UV signal is automatically normalized, which helps when comparing data from different systems.

Monitoring with multiple wavelengths can be used to detect contaminants, specifically labeled proteins, or target molecules that do not absorb light at 280 nm. To demonstrate this, molecular weight standards were monitored at 214, 280, and 340 nm wavelengths. Detection at 214 nm reveals peptide bonds of all proteins and may be useful if the concentration and extinction coefficient at 280 nm is low for the target protein. Ferritin, a multimeric iron-storage protein, showed stronger absorbance at 340 nm than the other proteins due to the high number of ferric ions in the center of the molecule (Fig 3).

Both UV monitor U9-L and UV monitor U9-M can be combined with a second UV monitor U9-L to give increased application capabilities such as multistep applications or when using small and large flow cells simultaneously to detect both low and high protein concentrations.

Column: Superdex™ 200 10/300 GL
Sample: Molecular weight standards for gel filtration
Sample volume: 100 µL
Eluent: PBS (10 mM sodium phosphate, 140 mM NaCl, 2.7 mM KCl, pH 7.4)
Flow rate: 0.5 mL/min
System: AKTA pure 25

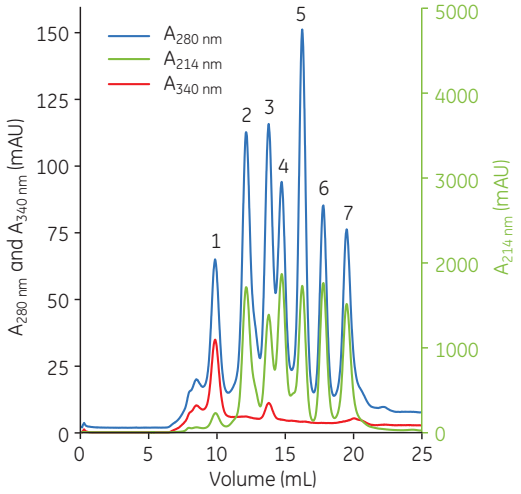


Fig 3. Gel filtration (GF) with multiwavelength detection (214, 280, and 340 nm) of proteins using ÄKTA pure with UV monitor U9-M. The column used was Superdex 200 10/300 GL. The peaks observed on the chromatogram are 1) ferritin (M_r 440 000), 2) aldolase (M_r 158 000), 3) conalbumin (M_r 75 000), 4) ovalbumin (M_r 44 000), 5) carbonic anhydrase (M_r 29 000), 6) ribonuclease A (M_r 13 700), and 7) aprotinin (M_r 6500).

Conductivity monitor

The conductivity monitor measures conductivity of buffer and samples for online monitoring of the true gradient. An integrated temperature sensor corrects for variations in conductivity due to the temperature. The conductivity monitor has a broad reading range and is therefore able to monitor conductivity in all different chromatographic techniques.

ÄKTA pure optional modules for enhanced automation

Sample application options

The optional sample pump (Fig 4) is designed to allow automatic sample application directly to a column or indirectly via a sample loop or Superloop. Using the sample pump saves time by eliminating laborious sample application steps and is especially useful when handling large sample volumes. The pump consists of two pump heads and is based on the same pump principle as the system pumps. Pump purging and air removal can easily be performed automatically. The sample pump is equipped with a pressure sensor for control of the sample flow rate to protect the column while preventing pressure stops and minimizing the time for sample loading. Using the sample pump, samples can be loaded at flow rates of up to 50 mL/min (Sample pump S9) or up to 150 mL/min (Sample pump S9H).

The optional sample inlet valve, V9-IS or V9H-IS, is intended to be used with the sample pump. Inlet valve allows fast, automatic loading of up to 7 different samples. The integrated air sensor enables complete sample application without the need to preprogram the sample volume. The valve has seven sample inlet positions plus a dedicated buffer inlet for filling the sample pump with solution before the sample is introduced and for washing out the valve and pump between runs. During sample application, the air sensor detects when sample has been completely loaded so that the method can continue to the next step without air being introduced into the flow path or column.

Fig 4. ÄKTA pure sample pump.

Buffer selection

ÄKTA pure can be equipped with two different types of inlet valves that allow selection of buffers and wash solutions. Valves with multiple inlets enable cleaning reagents to be permanently online, which means that columns and system can be cleaned conveniently at regular intervals.

Inlet selection valve, V9-IAB or V9H-IAB, comprises two A and two B inlet positions in a single valve offering a convenient solution for automation of buffer application and post-run cleaning of columns and system when performing basic chromatography. Any A inlet can be combined with any B inlet to generate gradients.

The inlet automation valves A and B provide up to 2 × 7 inlets. Multiple inlets enable automatic screening of buffer and reagent conditions. Each of the inlet automation valves is equipped with an integrated air sensor, which helps in excluding air from the system. If air is detected, the system can be paused so that the air can be purged before it enters the flow path.

Column control

A column valve can be connected to the system and used to control the flow to the column. ÄKTA pure can be equipped with one of two different column valves.

Column control valve, V9-Cs or V9H-Cs, allows connection of one column and has an integrated bypass function, which enables washing of the system without the need to remove the column. The column control valve also allows reverse flow through the column, for fast and effective elution of strongly bound proteins, sharper bands, and a concentrated target molecule eluent.

Column selection valve, V9-C or V9H-C, also has the integrated bypass and reverse-flow functions. Connection of up to five columns for automatic column switching is possible using this valve. Connection of multiple columns minimizes manual intervention and reduces further the risk of introducing air into the column.

The column selection valve has two integrated pressure sensors: the first sensor measures pressure before the column, enabling protection of the column hardware while the second measures the pressure after the column. The pressure drop over the column (Δp) is calculated by measuring the difference between the two pressure readings and can be used to protect the packed media bed (Fig 5).

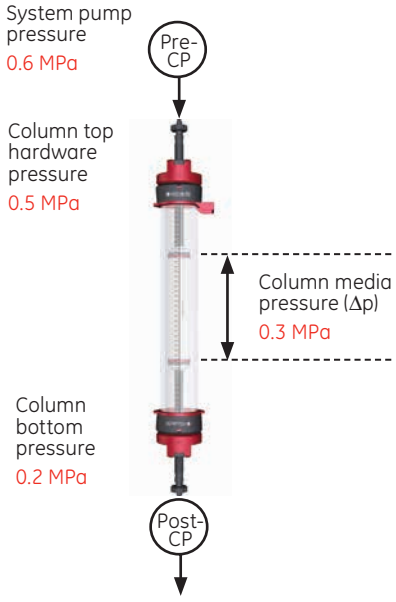


Fig 5. For increased operational safety, the column selection valve enables continuous measurement of precolumn (Pre-CP) and post-column pressure (Post-CP) during runs. The pressure difference over the packed media bed (Δp) is calculated from the two signals.

The flexibility of the column selection valve for connection of up to five columns was demonstrated in a column scouting study using columns for hydrophobic interaction chromatography (HIC). Five columns from HiTrap™ HIC Selection Kit were connected to ÄKTA pure and used for column scouting for optimization of purification conditions of S-aminotransaminase in clarified *E. coli* extract. UV monitor U9-M was used for detection of the protein at two wavelengths. Chromatograms of the five separate HIC runs are shown in Figure 6. Eluted fractions were analyzed using GF and SDS-PAGE (data not shown).

The A_{420} signal specifically monitors the target protein. The columns giving the sharpest and most symmetrical peaks at A_{420}, as well as the highest possible purity, were selected for subsequent optimization and scale-up experiments. HiTrap Phenyl FF (high sub) 1 mL and HiTrap Butyl FF 1 mL gave the most promising results under the conditions used, and HiTrap Phenyl FF (high sub) 1 mL was selected for further optimization in this case.

Columns:	Five columns from HiTrap HIC Selection Kit
Sample:	Supernatant after precipitation with 2 M ammonium sulfate (AS) at room temperature of extract of *E. coli* expressing S-aminotransaminase (adjusted to 1.5 M AS)
Sample volume:	2 mL
Buffer A:	1.5 M ammonium sulfate, 50 mM sodium phosphate, pH 7.0
Buffer B:	50 mM sodium phosphate, pH 7.0
Flow rate:	1 mL/min
UV cell:	10 mm
System:	AKTA pure 25 equipped with Column selection valve V9-C and Loop valve V9-L

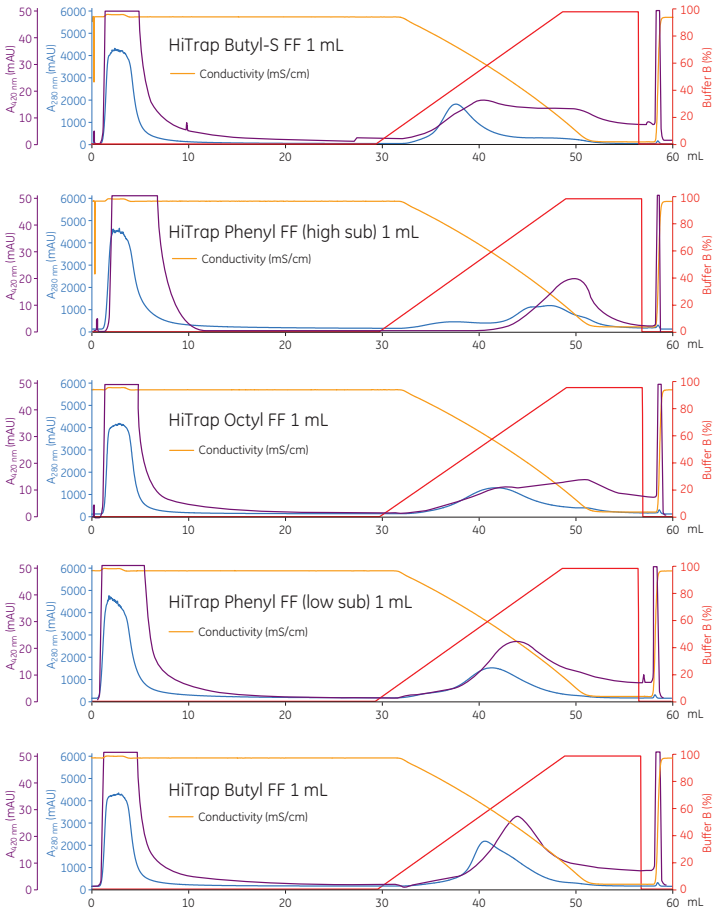


Fig 6. Column scouting for purification of S-aminotransaminase expressed in *E. coli*. Column selection valve V9-C allowed the connection of five HiTrap HIC columns to ÄKTA pure for this evaluation. UV monitor U9-M was used for multiwavelength detection. From this scouting, HiTrap Phenyl FF (high sub) 1 mL was selected for use in further scale-up studies.

pH monitoring

An optional pH valve with an integrated pH electrode (not included) enables in-line pH monitoring during the run. The pH monitor is easily calibrated by injection of calibration buffer directly into the valve with the pH electrode mounted. A flow restrictor is connected to the pH valve and can be automatically included in the flow path to generate a back pressure that prevents the formation of air bubbles in the UV flow cell. The pH valve is used to direct the flow to the pH electrode and flow restrictor, or alternatively, to bypass one or both. Bypassing the pH electrode means that it can be stored and kept in place on the valve at all times.

Outlet valves

Two different valve options are available to direct the flow to the fraction collector, waste, or other outlet ports. Outlet control valve, V9-Os or V9H-Os, allows connection of one or two fraction collectors. If only one is connected, the other port can be used for outlet fractionation, for example to collect flowthrough. Outlet fractionation valve, V9-O or V9H-O, enables connection of up to two fraction collectors, and up to 10 available outlets allow collection of large fractions.

Fraction collection

ÄKTA pure can be equipped with the Round fraction collector F9-R (Fig 7) or with the Flexible fraction collector F9-C (Fig 8). Both fraction collectors are controlled through UNICORN control software. Fraction collection can be based on time, volume, or automatic peak recognition. Automatic peak recognition minimizes cross-contamination and unwanted eluent can be diverted to the waste. For increased capacity, two units of fraction collector F9-R or one F9-R and one F9-C can be connected together.

Fraction collector F9-R provides a basic option with high capacity. A variety of racks is available to allow the use of 3, 8, 15, and 50 mL tubes. To minimize spillage, the Drop sync function can be used for flow rates up to 2 mL/min. Drop sync minimizes spillage by timing fraction changes between drops.

Fig 7. Fraction collector F9-R allows collection in 3, 8, 15, or 50 mL tubes.

Fraction collector F9-C provides flexibility, high capacity, and security. The fraction collector is equipped with a variety of cassettes that can hold tubes (3, 8, 15, and 50 mL) as well as deep well plates (24-, 48-, and 96-well), which means that samples can be collected in any format needed. Six cassettes can be loaded into the fraction collector in any combination that fits the user's needs (Fig 8). As an alternative to using six cassettes, loading capacity can be maximized by using a large tube rack for 50 mL tubes or a bottle rack for 250 mL bottles. Upon loading, the type of cassette is automatically detected by a sensor and the tube/bottle configuration is confirmed, eliminating mistakes in sample handling. Cassettes designed for tubes are equipped with a function that locks tubes into place when discarding liquid waste. Later, the tubes can be easily unlocked and discarded.

The cassettes can also be used for convenient storage of fractions or holders for sample tubes and are easy to handle and clean. The fraction collector is covered, protecting samples from dust contamination. The top of the fraction collector can be used for placement of accessories and equipment.

Fig 8. Fraction collector F9-C holds cassettes for a variety of tubes from 3 to 50 mL as well as 24-, 48-, and 96-deep-well plates.

Fraction collector F9-C has two beneficial features that minimize cross-contamination and spillage during fraction collection. Drop sync can be used for flow rates up to 2 mL/min and minimizes spillage by timing fraction changes to occur between drops. At higher flow rates, the Accumulator function provides spillage-free fractionation without sample-loss up to 150 mL/min. The system can automatically change between the two modes for optimal performance.

Additional module options

ÄKTA pure is a fully modular system that can be further expanded to increase system capability and productivity. Due to the accessibility and design of the modules, they are easily changed, which allows quick and efficient customization.

Versatile valve, V9-V or V9H-V, is a general four-position valve that can be used to tailor the system to specific tasks, for example, for multistep purification schemes (for more information, see Cue Card *Two-step purification with ÄKTA pure using loop collection*, code no. 29-0908-05). Up to four versatile valves can be connected to the system.

Mixer bypass valve, V9-M or V9H-M, is used for bypassing the mixer if samples are loaded through the system pump.

Loop valve, V9-L or V9H-L, allows the use of up to five loops and can be used for collection of intermediate fractions when performing multistep purification or for automated purification of up to five different samples. The loop valve can, for example, also be used for holding reagents or different samples.

Up to two extra eight-position inlet valves can be deployed to expand on buffer and sample inlet capacity. Up to four additional air sensors can be placed in the flow path to enhance security, for example, before the inlet valves or before the column.

I/O-box E9 provides a means of connecting external interfacing equipment such as detectors. I/O-box E9 receives analog or digital signals from, or transfers analog or digital signals to external equipment that needs to be incorporated in the system. Two I/O-box E9 units can be connected to ÄKTA pure.

A list of available additional valves and other options is found in Ordering information.

UNICORN 6 control software

UNICORN control software is used on all ÄKTA systems and can be used from lab scale up to full-scale production. UNICORN software contains the tools needed for beginners and experienced users to perform all types of chromatography, from setting up and running a method to evaluating the data.

Operation of ÄKTA pure is controlled by UNICORN 6 system control software, which provides built-in knowledge for planning, controlling, and analyzing the system and chromatography results. For greater convenience, licenses enabling remote control of ÄKTA pure are also available.

Key features of UNICORN 6 are:

- Intuitive software design: simple, intuitive, and flexible method creation using predefined phases (steps)
- Database storage: robust data storage allows easy access to data, data security, and data integrity
- Interactive process picture: shows the current open flow path and offers an intuitive way to control the run manually
- Column logbook (optional): valuable tool to keep track of individual column and run data for traceability and operational security
- DoE (optional): integrated tool for experimental design provides more precise information in fewer experiments for cost- and time-efficient method development

Method Editor

The Method Editor contains all the instructions used for controlling the chromatographic run. In UNICORN 6, the Method Editor has a user friendly graphical interface for easy viewing and editing of the method and run properties. Figure 9 shows a screenshot of the Method Editor with customizable panes that provide a comprehensive overview of the run.

In UNICORN 6, methods are built by using phases. Each phase reflects a step in the chromatography run, such as equilibration, sample application, or wash. The Method Editor contains predefined methods for different chromatography techniques and maintenance procedures, as well as a library of predefined phases for creating or editing your own methods.

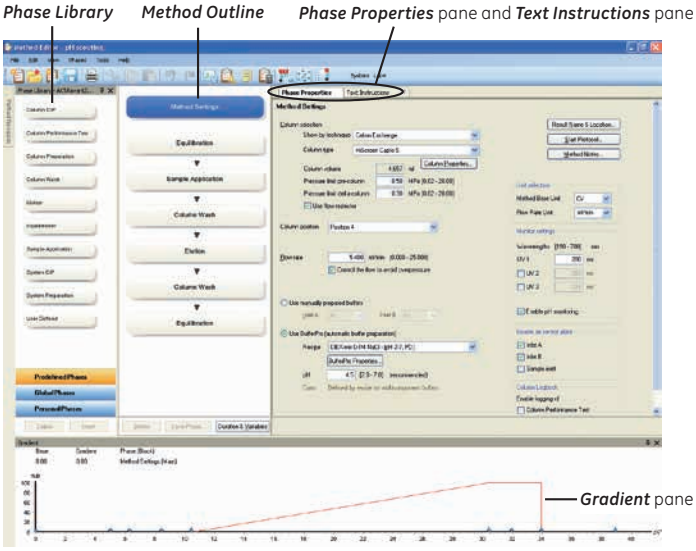


Fig 9. Method Editor has customizable panes that give a comprehensive overview of the method.

A method is created or edited simply by dragging and dropping phases from the Phase Library into the Method Outline. In the Phase Properties pane, specific run parameters are set, and these settings are automatically programmed in the Text Instructions pane. When selecting a column type, parameter settings for that column type (e.g., default flow rates and pressure limits) are automatically set.

The Phase Properties pane allows for easy editing of the method, while more advanced users may program the method directly in the Text Instructions pane. For quick editing, the toolbar contains useful icons such as Undo, Redo, and Save.

System Control

The System Control module is used to start, view, and control a method run. The module consists of three panes that provide an overview of the status of the run. The Run Data pane presents current data in numerical values, while the Chromatogram pane illustrates data as curves during the entire method run. The Process Picture pane displays the current flow path during the run and can be used to control the run (Fig 10). Color indication incorporated in the process picture shows the current open flow path with flow, closed flow path, or open flow path without flow. Real-time data from monitors are also displayed in the process picture pane.

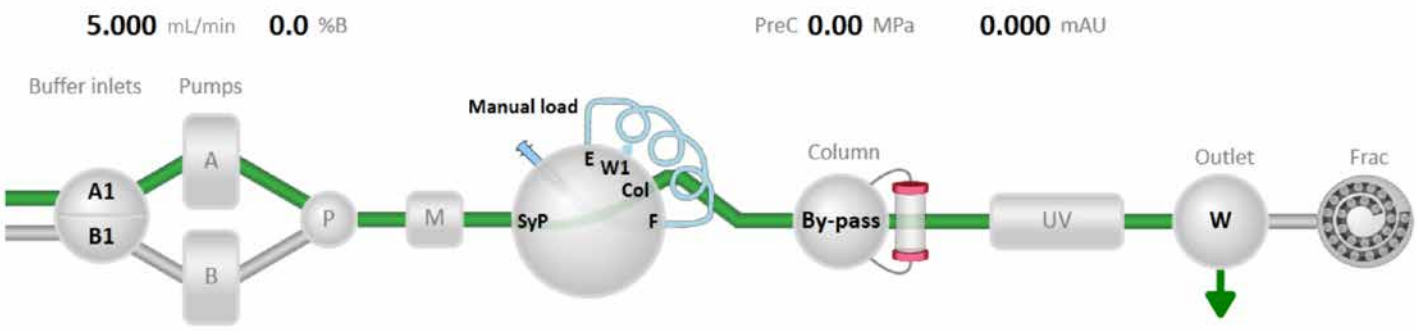


Fig 10. The process picture in UNICORN 6 shows currently active and inactive flow paths, and provides a fast and easy way to control the system.

Column logbook

To increase operational safety, an optional feature of the software is the Column logbook. The practical Column logbook tool keeps track of important run data related to individual columns to provide traceability and operational security. Many prepacked columns from GE Healthcare are barcode-labeled, and individual columns are identified using a 2-D barcode scanner, or the information may be entered manually into UNICORN 6. UniTag sticker labels, with preprinted 2-D barcodes, are available for labeling columns that are not already barcode-labeled (e.g., empty columns).

By tracking individual columns, information regarding run data such as total number of runs and maximum pressures is recorded for each run. Notification limits can be set, for instance, to define the number of times the column may be run between cleanings, and the user is notified when it is time for column maintenance. In the Column logbook, clicking on the Column History icon for a particular column provides a list of all runs that have been performed with that specific column.

Design of Experiments (DoE) functionality

DoE functionality can be added as an option. DoE is a powerful tool for method optimization. DoE provides an efficient and structured approach where selected parameters are varied simultaneously so that a large data set can be obtained from few experiments (Fig 11). Since DoE is integrated seamlessly in UNICORN 6, scouting methods are automatically generated from DoE schemes, allowing for fast and efficient method optimization.

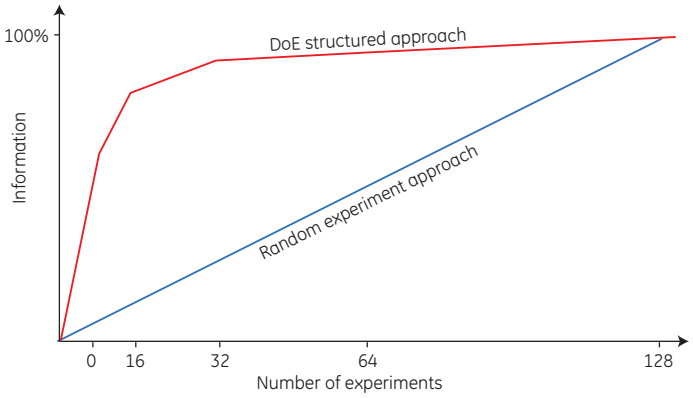


Fig 11. The DoE module, which is optional in UNICORN 6, provides an efficient approach to optimization, capturing more information in fewer experiments.

Accessories

ÄKTA pure accessories include column holders and clamps for attaching columns, flasks, and tubing to the system (Fig 12). A selection of tubing kits allow a optimization of the flow path for various objectives and the connection of any laboratory-scale column from GE Healthcare.

Fig 12. ÄKTA pure accessories include holders and clamps for attaching columns, flasks, and tubing to the system.

* To use as an air sensor holder, the adapter 28-9563-42 is also needed
† To be used to attach, for example, fraction collector cassettes on the side of the system

Prepacked columns complete the package

GE Healthcare offers an extensive range of prepacked columns for purification, from microgram levels to hundreds of milligrams of target protein and for almost every chromatography technique (Fig 13). The range includes HiTrap, HiPrep™, HiScreen™, and HiLoad™ columns for preparative chromatography. Tricorn™ columns are also available for high-resolution semipreparative purifications at microgram scale as well as for protein characterization. In addition to prepacked columns, empty columns for packing with chromatography media of your choice are available.

Columns for microgram-scale purification

Tricorn GL and PE columns are high-performance columns prepacked with media for a variety of chromatography techniques (Fig 13). The column design allows even distribution of liquid eluent over the entire column cross-section, which enables high-resolution purification at micro- and milligram scale. Tricorn GL columns are manufactured in glass to facilitate visual inspection of the media bed while the tube and filter of PE columns are designed to withstand greater pressure.

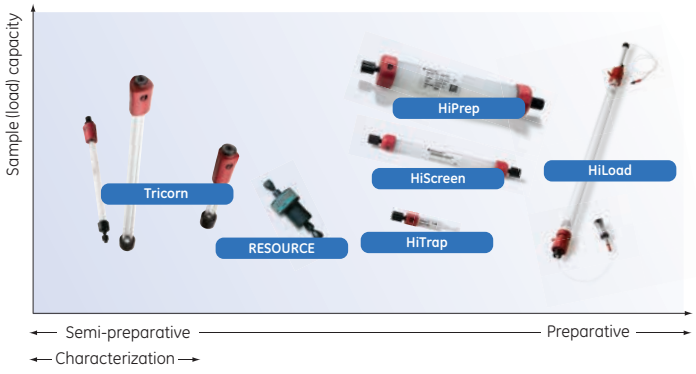
Columns for milligram-scale purification

HiTrap 1 and 5 mL columns are prepacked with a wide range of media for purification using various chromatography techniques (Fig 13). The columns can be connected in series for greater capacity. Further scale-up can be achieved with HiPrep 20 mL columns.

RESOURCE™ columns are designed for high-resolution purification of proteins at high flow rates. The columns are prepacked with SOURCE™ media that have high particle size uniformity and stability to allow high flow rates at low back pressure.

HiScreen columns are prepacked with a wide range of robust BioProcess™ media to allow repeated use with highly reproducible results. Designed for scalable method optimization, HiScreen columns have a 10 cm bed height and can easily be connected in series to achieve a 20 cm bed height.

HiLoad columns are prepacked glass columns with Superdex prep grade media designed for high-resolution GF applications.



Tricorn	RESOURCE	HiTrap	HiScreen	HiPrep	HiLoad
High quality and high resolution (GF, IEX, HIC)	Fast purifications with good resolution (IEX, HIC, RPC)	Easy to use for a broad range of applications (AC, DS, IEX, HIC)	Optimized for method and process development (AC, IEX, HIC)	Fast and easy scale-up (AC, DS, GF, IEX, HIC)	Preparative gel filtration with high resolution (GF)

Fig 13. Columns for use with ÄKTA pure system for different scales of purification. AC = affinity chromatography, DS = desalting, GF = gel filtration, IEX = ion-exchange chromatography, HIC = hydrophobic interaction chromatography, RPC = reversed-phase chromatography.

Pack your own columns for gram-scale purification

The column packing instruction in ÄKTA pure enables convenient column packing at constant pressure and high flow rates. Both A and B pumps are used to generate the flow, making it possible to set flow rate up to 50 mL/min and 300 mL/min for ÄKTA pure 25 and ÄKTA pure 150, respectively.

Several empty column types are avaliable. Which empty column that is best suited will depend upon media type and running conditions (i.e. flow, pressure etc). HiScale™ empty columns are developed for standard liquid chromatography, optimized for process development and preparative protein purification. The columns are designed to withstand high pressures and high flow rates making them compatible with modern BioProcess chromatography media such as MabSelect™ and Capto™ media.

XK columns are user friendly and robust columns for standard protein purification. The columns are designed for liquid chromatography at low to medium pressure.

System specifications

Control system	UNICORN 6 (version 6.3 or later)
Dimensions (W × H × D)	535 × 630 × 470 mm
Weight (excluding computer, sample pump, fraction collector)	Up to 53 kg
Power supply	100–240 V, ~50–60 Hz
Power consumption	300 VA (typical), 25 VA (power-save)
Enclosure protective class	IP 21

System pump

Pump type	Piston pump, metering type
Flow rate setting	ÄKTA pure 25: 0.001 to 25 mL/min (up to 50 mL/min during column packing) ÄKTA pure 150: 0.01 to 150 mL/min (up to 300 mL/min during column packing)
Flow rate specifications	ÄKTA pure 25: Accuracy: ± 1.2% Precision: RSD < 0.5% (conditions: 0.25 to 25 mL/min, < 3 MPa, 0.8 to 2 cP) ÄKTA pure 150: Accuracy: ± 1.5% Precision: RSD < 0.5% (conditions: 1.0 to 150 mL/min, < 3 MPa, 0.8 to 2 cP)
Pressure range	ÄKTA pure 25: 0 to 20 MPa (2900 psi) ÄKTA pure 150: 0 to 5 MPa (725 psi)
Viscosity range	ÄKTA pure 25: 0.35 to 10 cP (5 cP above 12.5 mL/min) ÄKTA pure 150: 0.35 to 5 cP

Sample pump

Pump type	Piston pump, metering type
Dimensions (W × H × D)	215 × 210 × 370 mm
Weight	11 kg
Flow rate setting	ÄKTA pure 25: 0.001 to 50 mL/min ÄKTA pure 150: 0.01 to 150 mL/min
Flow rate specifications	ÄKTA pure 25: Accuracy: ± 2% Precision: RSD < 0.5% (conditions: 0.25 to 50 mL/min, < 3 MPa, 0.8 to 3 cP) ÄKTA pure 150: Accuracy: ± 2% Precision: RSD < 0.5% (conditions: 1.0 to 150 mL/min, < 3 MPa, 0.8 to 3 cP)
Pressure range	ÄKTA pure 25: 0 to 10 MPa (1450 psi) ÄKTA pure 150: 0 to 5 MPa (725 psi)
Viscosity range	0.7 to 10 cP

Mixer

Mixing principle	Chamber with a magnetic stirrer
Mixer volume	ÄKTA pure 25: 0.6, 1.4 (mounted on system), or 5 mL ÄKTA pure 150: 1.4 (mounted on system), 5 mL (included with system), or 15 mL
Gradient flow rate range	ÄKTA pure 25: 0.1 to 25 mL/min ÄKTA pure 150: 0.5 to 150 mL/min
Gradient composition accuracy	ÄKTA pure 25: ± 0.6% (conditions: 5 to 95% B, 0.6 to 25 mL/min, 0.2 to 2 MPa, 0.8 to 2 cP) ÄKTA pure 150: ± 0.8% (conditions: 5 to 95% B, 2 to 150 mL/min, 0.2 to 2 MPa, 0.8 to 2 cP)

Valves

Type	Rotary valves
Number of valves	Up to 12
Functions	Standard: injection valve Options: inlet selection, mixer by-pass, loop selection, column selection, pH, outlet, versatile

Pressure sensors

Placement of sensors	Standard: after system pump Options: after sample pump, precolumn, post-column
Range	0 to 20 MPa (2900 psi)
Accuracy	± 0.02 MPa or ± 2%, whichever is greater

Module options

Inlet valves

Inlet A	1, 2, or 7 inlets
Inlet B	1, 2, or 7 inlets
Sample inlet	0, 1, or 7 inlets
Additional inlets	Up to 16

UV monitors

Number of monitors	Up to 2 (two U9-L or one U9-L and one U9-M)
Wavelength range	UV monitor U9-L: 280 nm UV monitor U9-M: 190 to 700 nm in steps of 1 nm, up to 3 simultaneous wavelengths
Absorbance range	-6 to +6 AU
Linearity	Within ± 2% at 0 to 2 AU
Operating pressure	0 to 2 MPa (290 psi)
Flow cells: U9-L	Standard: Optical path length 2 mm Cell volume 2 µL Options: Optical path length 5 mm Cell volume 6 µL
Flow cells: U9-M	Standard: Optical path length 2 mm Cell volume 2 µL Options: Optical path length 10 mm Cell volume 8 µL Optical path length 0.5 mm Cell volume 1 µL

Conductivity monitor, C9n

Conductivity reading range	0.01 mS/cm to 999.99 mS/cm
Accuracy	± 0.01 mS/cm or ± 2%, whichever is greater (within 0.3 to 300 mS/cm)
Operating pressure	0 to 5 MPa (725 psi)
Flow cell volume	22 µL
Temperature monitor range	0°C to 99°C
Temperature monitor accuracy	± 1.5°C within 4°C to 45°C

pH monitor, V9-pH

pH reading range	0 to 14
Accuracy	± 0.1 pH unit within pH 2 to 12
Operating pressure	0 to 0.5 MPa (72 psi)
Flow cell volume	ÄKTA pure 25: 76 µL ÄKTA pure 150: 129 µL

Round fraction collector, F9-R

Number of F9-R	Up to 2 (two Round fraction collector F9-R or one F9-R and one Flexible fraction collector, F9-C)
Number of fractions	Up to 175 per fraction collector
Vessels	175 (3 mL tubes) 85 (8 or 15 mL tubes) 40 (50 mL tubes)
Fraction volumes	0.1 to 50 mL
Spillage-free mode	Drop sync
Flammable liquids	Yes
Dimensions (W × H × D)	320 × 250 × 400 mm
Weight	5 kg
Delay volume (UV – dispenser head)*	ÄKTA pure 25: 205 µL (86 µL with optional tubing kit, i.d. 0.25 mm) ÄKTA pure 150: 473 µL (278 µL with optional tubing kit, i.d. 0.5 mm)

Flexible fraction collector, F9-C

Number of F9-C	1 (if needed add one Round fraction collector, F9-R)
Number of fractions	Up to 576
Number of cassettes†	6
Number of cassette trays†	1
Vessel types	Tubes per cassette: 40 (3 mL tubes), total per tray 240 24 (8 mL tubes), total per tray 144 15 (15 mL tubes), total per tray 90 6 (50 mL tubes), total per tray 36 Plates per cassette: 1 deep well plate (24, 48 or 96 wells), 6 plates per tray Tubes per cassette tray: 55 (50 mL tubes) Bottles per cassette tray: 18 (250 mL bottles of squared shape)
Fraction volumes	0.1 to 250 mL
Spillage-free mode	Drop sync, Accumulator, or Automatic
Flammable liquids	No
Dimensions (W × H × D)	390 × 320 × 585 mm
Weight	21 kg
Delay volume (UV – dispenser head)*	ÄKTA pure 25: 435 µL (214 µL with optional tubing kit, i.d. 0.25 mm) ÄKTA pure 150: 876 µL (508 µL with optional tubing kit, i.d. 0.5 mm)

Outlet valves

Number of outlets	Valve V9-Os or V9H-Os: 3 (waste, fraction collector, 1 outlet position) Valve V9-O or V9H-O: 12 (waste, fraction collector, 10 outlet positions)
Fraction volumes	0.01 to 100 000 mL
Delay volume (UV – outlet valve)	ÄKTA pure 25: 125 µL (66 µL with optional tubing kit, i.d. 0.25 mm) ÄKTA pure 150: 296 µL (245 µL with optional tubing kit, i.d. 0.5 mm)

Air sensors

Number of sensors	Up to 7
Placement of built-in sensors	Inlet valve V9-IA, Inlet valve V9-IB, Sample inlet valve V9-IS
Placement of additional sensors	After the injection valve sensors Before the system pumps Before the sample pump
Sensing principle	Ultrasonic

I/O-box E9

Number of I/O boxes	2
Number of ports per box	2 analog in, 2 analog out 4 digital in, 4 digital out
Analog range	In ± 2 V Out ± 1 V

* The delay volume will change if a different tubing length between the system and the fraction collector is used.

† The fraction collector can hold either up to six cassettes or one cassette tray.

Ordering information

Product	Code number
ÄKTA pure 25 L	29-0182-24
ÄKTA pure 25 M	29-0182-26
ÄKTA pure 25 L1 (V9-IAB, V9-Os)	29-0182-25
ÄKTA pure 25 M1 (V9-IAB, V9-Os)	29-0182-27
ÄKTA pure 25 M2 (V9-IA, V9-IB, V9-C, V9-O)	29-0182-28
ÄKTA pure 150 L	29-0466-65
ÄKTA pure 150 M	29-0466-94
ÄKTA pure 150 M3 (V9H-IA, V9H-C, V9H-O)	29-0466-97
ÄKTA pure User Manual, printed copy (digital included)	29-2827-26
UNICORN 6.4 workstation License	29-0561-02
UNICORN 6.4 Remote License without DVD	29-0670-44
UNICORN 6.4 Dry License without DVD	29-0670-45
UNICORN 6.4 DoE concurrent license	29-0670-46
UNICORN 6.4 Column logbook, network license	29-0670-47
UNICORN 6.4 DVD package	29-0561-00
UNICORN 6.4 Manual package	29-0670-74

System modules and accessories

Mixer

Mixer chamber 0.6 mL	28-9561-86
Mixer chamber 1.4 mL (incl. with all systems)	28-9562-25
Mixer chamber 5 mL (incl. with ÄKTA pure 150)	28-9562-46
Mixer chamber 15 mL	28-9803-09
Online filter kit	18-1027-11
O-ring 13.1 × 1.6 mm, high resistance	29-0113-26
O-ring 13.1 × 1.6 mm (for Mixer chamber 0.6, 1.5 and 5 mL)	28-9535-45
O-ring 22.1 × 1.6 mm (for Mixer chamber 15 mL)	28-9818-57

Valves ("valve kit" include necessary tubing)*

	ÄKTA pure 25	ÄKTA pure 150
Sample inlet valve kit	(V9-IS) 29-0277-46	(V9H-IS) 29-0509-43
Inlet valve kit A	(V9-IA) 29-0122-63	(V9H-IA) 29-0509-45
Inlet valve kit B	(V9-IB) 29-0123-70	(V9H-IB) 29-0509-46
Inlet valve kit AB	(V9-IAB) 29-0113-57	(V9H-IAB) 29-0896-52
Inlet valve X1	(V9-X1) 28-9572-27	(V9H-X1) 28-9793-26
Inlet valve X2	(V9-X2) 28-9572-34	(V9H-X2) 28-9793-28
Mixer valve kit	(V9-M) 29-0113-54	(V9H-M) 29-0906-92
Loop valve kit	(V9-L) 29-0113-58	(V9H-L) 29-0906-89
Column valve	(V9-Cs) 29-0113-55	(V9H-Cs) 29-0906-93
Column selection valve	(V9-C) 29-0113-67	(V9H-C) 29-0509-51
pH valve kit	(V9-pH) 29-0113-59	(V9H-pH) 29-0516-84
Versatile valve	(V9-V) 29-0113-53	(V9H-V) 29-0906-91
Outlet valve kit (10 outlets)	(V9-O) 29-0122-61	(V9H-O) 29-0509-49
Outlet valve kit (1 outlet)	(V9-Os) 29-0113-56	(V9H-Os) 29-0906-94

* The valves for ÄKTA pure 25 and ÄKTA pure 150 are compatible with both systems but for optimal performance, the specific valve type should be used.

UV monitor	Code number
UV monitor U9-L	29-0113-60
UV flow cell U9-0.5, 0.5 mm for U9-M	28-9793-86
UV flow cell U9-2, 2 mm for U9-M (incl. in system with U9-M)	28-9793-80
UV flow cell U9-10, 10 mm for U9-M	28-9563-78
UV flow cell 2 mm for U9-L (incl. with U9-L)	29-0113-25
UV flow cell 5 mm for U9-L	18-1128-24

Sample pump

Sample pump S9	29-0277-45
Sample pump S9H	29-0505-93

pH and conductivity monitors

pH electrode	28-9542-15
O-ring 5.3 × 2.4 mm (for pH electrode)	28-9564-97
Conductivity monitor C9	29-0113-63

Injection valve accessories

Sample loop 10 µL	18-1120-39
Sample loop 100 µL	18-1113-98
Sample loop 500 µL (incl. with all systems)	18-1113-99
Sample loop 1 mL	18-1114-01
Sample loop 2 mL	18-1114-02
Sample loop 10 mL	18-1161-24
Superloop 10 mL	19-7585-01
Superloop 50 mL	18-1113-82
Superloop 150 mL	18-1023-85

Fraction collector F9-R

Fraction collector F9-R	29-0113-62
Tube Rack with 175 positions for 12 mm vials, bowl, tube support, holder and guide	19-8684-03
Tube Rack with, 95 positions for 10–18 mm vials	18-3050-03
Tube Rack with 40 positions for 30 mm vials, bowl, tube support, holder and guide	18-1124-67

Fraction collector F9-C

Fraction collector F9-C	29-0277-43
Cassette tray, holds up to six cassettes	28-9542-09
Cassette, holds six 50 mL tubes (2-pack)	28-9564-02
Cassette, holds fifteen 15 mL tubes (2-pack)	28-9564-04
Cassette, holds twenty-four 8 mL tubes (2-pack)	28-9564-25
Cassette, holds forty 3 mL tubes (2-pack)	28-9564-27
Cassette, holds one 96-, 48-, or 24- deep-well plate (2-pack)	28-9542-12
Rack, holds fifty-five 50 mL tubes	28-9803-19
Rack, holds eighteen 250 mL bottles	28-9818-73

Additional air sensors

Air sensor L9-1.2 mm	28-9565-02
Air sensor L9-1.5 mm	28-9565-00
Adapter for air sensor	28-9563-42

I/O-box E9

I/O-box E9	29-0113-61

Barcode labels and scanner

UniTag (1 sheet with 108 labels)	28-9564-91
Barcode scanner 2-D with USB	28-9564-52

Tubing kits	Code number	
	ÄKTA pure 25	ÄKTA pure 150
Tubing kit i.d. 0.25 mm	29-0113-28	–
Tubing kit i.d. 0.5 mm (std. ÄKTA pure 25)	29-0113-27	29-0516-69
Tubing kit i.d. 0.75 mm (std. ÄKTA pure 150)	29-0113-29	29-0482-42
Tubing kit i.d. 1.0 mm (incl. ÄKTA pure 150)	29-0324-26	29-0324-26
Tubing kit for sample inlet valve (7 inlets)	29-0353-31	29-0511-66
Sample tubing kit for 7 inlets, i.d. 0.75 mm	28-9572-17	28-9572-17
Inlet tubing kit for inlet valve IAB	29-0113-30	29-1064-97
Tubing kit for pH valve, standard	29-0113-31	29-0516-74
Tubing kit for inlet valve A (7 ports)	29-0113-32	29-0511-97
Tubing kit for inlet valve B (7 ports)	29-0113-33	29-0511-89
Tubing kit for outlet fractionation (10 outlets)	29-0113-34	29-0486-11
System and sample pump rinse tubing kit	29-0113-48	29-0113-48

Cables	
Jumper D-SUB	29-0113-65
Jumper 1 IEC 1394 (F-type)	28-9564-89
External module cable, short	29-0124-74
External module cable, long	29-0113-66
2.5 m cable for F9-C or S9 (UniNet-9 D-type)	29-0324-25

Holders	
Column holder rod	28-9562-70
Tubing holder spool	28-9562-74
Tubing holder spool (i.d. 2.9 mm tubing) for ÄKTA pure 150	29-0142-83
Column and bottle holder o.d. 10–50 mm	28-9562-82
Tubing holder comb	28-9562-86
Flexible column holder for HiScreen columns	28-9562-95
Inlet filter holder Kit, ÄKTA	11-0004-07
Column clamp o.d. 10–21 mm	28-9563-19
Adapter for air sensor	28-9563-42
Bottle and airsensor holder*	28-9563-27
Tube holder (5-pack)	28-9543-29
Multipurpose holder	29-0113-49
Rail extension	29-0113-52
Loop holder with five 10 mL sample loops	29-0113-50
Screw lid kit, ÄKTA	11-0004-10

* To use as an airsensor holder the adapter 28-9563-42 is also needed.

Related literature

	Code number
UNICORN 6 control software, Data file	28-9573-46
Validation Support File UNICORN software, Data file	28-9626-50
ÄKTA pure: Protein purification your way, Brochure	29-0211-97
Purification of a miniature recombinant spidroin protein expressed in *E. coli* using ÄKTA pure system, Application note	29-0211-98
Purification and immobilization of a transaminase for the preparation of an enzyme bioreactor, Application note	29-0211-99
Prepacked chromatography columns for ÄKTA systems, Selection guide	28-9317-78
Two-step purification with ÄKTA pure using loop valve, Cue card	29-0908-05
Tandem two-step purification using ÄKTA pure, Cue card	29-0908-06
Connect Alias™ autosampler to ÄKTA pure, Instructions	29-0404-27
ÄKTA laboratory-scale chromatography systems, Instrument management handbook	29-0108-31
Design of experiments (DoE) in protein production and purification, Handbook	29-1038-50

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GE Healthcare Bio-Sciences AB
Björkgatan 30
751 84 Uppsala
Sweden

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GE Healthcare UK Limited
Amersham Place
Little Chalfont
Buckinghamshire, HP7 9NA
UK

GE Healthcare Europe, GmbH
Munzinger Strasse 5
D-79111 Freiburg
Germany

GE Healthcare Bio-Sciences Corp.
800 Centennial Avenue, P.O. Box 1327
Piscataway, NJ 08855-1327
USA

GE Healthcare Japan Corporation
Sanken Bldg., 3-25-1, Hyakunincho
Shinjuku-ku, Tokyo 169-0073
Japan

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